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L9: Entry 12 of 18 File: USPT 102 Apr 28, 1981

DOCUMENT-IDENTIFIER: US 4264448 A
TITLE: Method for bacteriological treatment of manure and high bod industrial wastes

BSPR:
The animal industry generally in the United States produces about 2 billion tons of animal manure a year. For example, such feedlots or feedyards may often contain as many as 2,000 to 50,000 head of cattle in a relatively small area. Typically, an average of 18 pounds (dry weight) per day of high value feed mixes must be fed to each animal for maintenance and to produce a daily average increase in weight of 11/2 to 23/4 pounds. This average animal voids approximately 6 pounds of dry weight per 24 hour period. Typically the manure is merely removed periodically from the confined areas and stockpiled, pressed into blocks and in some cases a small amount has been used on fields as humus. Animal wastes are thus accumulated in localized areas and become sources of air and water pollution. The amount of animal waste generated in the United States is about 10 times that of human waste and 70% of this animal waste is from cattle.

BSPR:
Thus, there exists the need to treat animal waste either as a soil extender without any environmental concerns or with modification as a feed for cattle.

BSPR:
My invention is broadly directed to a process for the treatment of manure, either raw or sterilized, from ruminants which treatment renders the manure acceptable either as an animal feed, as a fertilizer, or an environmentally acceptable landfill.

DEPR:
The reduction in bacteria (other than the bacteria used for the acid fermentation process), will be as described in my parent application. That is, after the acid fermentation, the coliform and total gram negative bacteria measurements will be negative. The addition of the lactobacilli and fermentable carbohydrate, whether or not the manure is sterilized prior to the acid fermentation, is sufficient over a predetermined period of time, say three to five days, to lower the pH sufficiently so that the undesirable bacteria is reduced to a level wherein the treated manure in this one-step process renders that treated manure usable either as a feed for animals with the addition of other nutrients or as a fertilizer or land fill.

DEPR:
Where L. plantarum is used, it may be grown and harvested in the manner as set forth in my parent application. The concentrations of bacteria added to the manure will range between about 10.sup.2 to 10.sup.3 cells/ml. Preferably the carbohydrate or the disaccharide lactose and the specific bacteria L. plantarum are used. The other species of the genus Lactobacillus alone or in combination are also suitable. The temperature range for growth is typically 5.degree.-53.degree. C. The lactobacilli are acidophillic with an optimal initial pH range of 5.5 to 5.8 and clearly grow at a pH of 5.0 or less. The complex nutritional requirements of lactobacilli for amino acids, peptides, nucleic acid derivatives, vitamins, salts, fatty acids or fatty acid esters appear to be present in typical manure. It has been found that additional fermentable carbohydrates, however, must be added to the manure for the pH to drop below 4.5. Any one of the following bacteria or combinations thereof may be used with my invention: L. acidophilus, L. bulgaricus, L. casei, L. coryniformis, L. delbruckii, L. helveticus, L. lactis, L. leichmannii, L. plantarum, L.

thermophilus, L. xylosus, L. brevis, L. buchneri, L. coprophilus, L. fermentum, L. viridenscens.

DEPR:
In a further alternative embodiment of the invention, the stabilized mass resulting from the above process (either preferred or alternative) may be further treated to produce ammonium lactate, a feed supplement for animals. After the bacteria population of the manure has been stabilized (pathogenic and gram negative bacteria have been killed by exposure to pH of 3.8 to 4.2) aqueous ammonium is bubbled through the manure forming ammonium lactate. The ammonium may be bubbled through in any conventional manner. During the process, the lactobacilli are digesting fermentable carbohydrates and producing lactic acid. The pH during this step is between 4.5 and 5.5. The pH is maintained in this range by controlling both the amount of ammonia and fermentable carbohydrate being added to the manure.

CLPR:
7. The method of claim 1 wherein the bacteria is selected from the group consisting of L. acidophilus, L. bulgaricus, L. casei, L. coryniformis, L. delbruckii, L. helveticus, L. lactis, L. leichmannii, L. plantarum, L. thermophilus, L. xylosus, L. brevis, L. buchneri, L. coprophilus, L. fermentum, L. viridescens.

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L. Kefir

L13: Entry 6 of 15 File: USPT 102(e) Sep 22, 1998

Filed 2/6/96

DOCUMENT-IDENTIFIER: US 5811289 A
TITLE: Process and apparatus for effecting a biological aerobic pretreatment of dairy industry effluent

ABPL:
This aerobic waste pretreatment process comprises inoculating a milk industry effluent with a mixture of bacteria and yeasts both classes of microorganisms capable of living and growing in symbiosis in the effluent, the population of the bacteria being, in most cases, several times greater than the population of the yeasts, maintaining the temperature and pH of the inoculated effluent between 0.degree. C. and 50.degree. C. and between 1.7 and 9, aerating the effluent while varying, if necessary, the pH at maximum rate of 1.5 pH units per minute and also, if required, modulating the aeration of the inoculated effluent at a maximum rate of 130 micromoles of oxygen per minute. A biomass is obtained which has a good nutritional value suitable for animal feed.

BSPR:
A corollary object consists in the production of a biomass with enhanced nutritional value usable as animal feed which would be a source of revenue for the dairy industry.

DEPR:
When the biomass produced according to the present invention is intended for animal feed, the appetency can be increased by developing aroma by the adjunction to the base strains of one, several or all of the following strains: Acetobacter aceti, Propionibacterium freundenreichii shermanii, Pseudomonas fragi, and Streptococcus lactis diacetylactis. Those species develop very moderately in effluent, either because of competition or because their growth is naturally slow, as is the case for example for Propionibacterium freudenreichii shermanii, or because the conditions of pH or aeration are not optimal for their growth.

DEPR:
When it comes out of the reactor, the liquor is submitted to liquid-solid phase separation by the method best suited to each case: decantation, centrifugation, filtration, etc. Thus the recovered biomass is either totally unloaded or partly recycled in the reactor in the case where a low input load operation is preferable. The excess biomass taken out of this pretreatment process is stored for further use in trophic chains, namely animal feed for bovids, crustaceans and the like.

DEPR:
The nitrogen in the form of a nitrogen compound such as ammonia or an ammonium salt is prepared as a solution within a vessel 13 and is injected by a dosage pump 14 into the hydrolysing apparatus 10 (or optionally directly into the reactor vessel 2). The liquor unloaded from the reactor vessel 2 is introduced into a phase separator 16 which is shown as a centrifugal apparatus but which could be any other type of phase separator. The pretreated water is unloaded by piping 17 whereas the separated biomass is partially recycled into the reactor vessel 2 by piping 18 and partially into the hydrolysing apparatus 10 by an optional piping 20. The excess biomass is stored in a storage reservoir 19 before being discharged as animal feed by a conduit 22. A heat exchanger coil 21 within vessel 2 serves to extract heat energy from vessel 2 to be used elsewhere or to cool the liquor.

DEPR:
The COD of the pretreated water is 1950 milligram per liter and the COD

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L9: Entry 9 of 18 File: USPT ~ 102/103 Sep 11, 1990

DOCUMENT-IDENTIFIER: US 4956295 A
TITLE: Stabilization of dried bacteria extended in particulate carriers

BSPR:
No satisfactory method has been known for producing consistent, reproducible stabilization of bacteria in dry carriers. From a practical standpoint, the problem has been further complicated by observations that greater instability, and greater uncertainty of results occur as the degree of dilution of the bacteria is increased. Yet this is exactly the type of formulation most desired for the large scale uses like those of L. plantarum and acidophilus referred to above. Prior art disclosures describe the survivability of lactic acid bacteria at low or room temperature. However, the amount of bacteria surviving extended storage periods is relatively small, especially at room temperature. None of the prior art disclosures previously described disclose methods to stabilize bacteria for extended periods of time while maintaining the viability of the bacteria, i.e., large number of the bacteria to function as inoculates of animal feed.

BSPR:
Concentrated cultures of lactic acid producing bacteria may be prepared by the method of U.S. Pat. No. 4,115,199. Tripolyphosphate and/or hexametaphosphate are added to the culture medium prior to separation of the cells by centrifugation. The resulting concentrates have usually been frozen with liquid nitrogen for use in manufacturing cheese or other dairy products. However, sufficient stabilization for distribution in a dry non-refrigerated form can be obtained by the method described in U.S. Pat. No. 3,897,307. The cell culture is adjusted to a pH favorable to the stability of the cells on drying, and chemical stabilizers are added comprising an ascorbate compound together with either a glutamate compound or an aspartate compound (viz. ascorbic acid with monosodium glutamate). The bacteria are then dried by a suitable procedure; viz. freeze-drying, spray drying or fluid bed drying. Drying to a low moisture content, such as 2.5 to 3.5% by weight, is desirable. Good stability is obtained when the product is packaged in moisture impervious containers. This permits non-refrigerated storage and distribution for uses such as home manufacture of yogurt. Such stabilization procedures, however, have not been adequate where the bacteria are mixed with relatively large amounts of particulate carriers to form highly extended bacterial admixtures, such as for addition to animal feeds or addition to silage materials.

DETL:
Lactobacillus bulgaricus Lactobacillus coryniformis Lactobacillus acidophilus subspec. coryniformis Lactobacillus helveticus Lactobacillus curvatus Lactobacillus bifudus Lactobacillus brevis Lactobacillus casei Lactobacillus buchneri Lactobacillus lactis Lactobacillus fermentum Lactobacillus plantarum Lactobacillus viridescens Lactobacillus delbrueckii Lactobacillus amylovorus Lactobacillus thermophilus Lactobacillus amylophilus Lactobacillus fermetii Lactobacillus pentosaceus

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L9: Entry 11 of 18 File: USPT May 27, 1986

DOCUMENT-IDENTIFIER: US 4591499 A
TITLE: Method for treatment and prevention of mastitis

DEPR:
Cultures of lactic acid producing bacteria have long been used in the manufacture of cheese, yogurt, buttermilk and other dairy products. Beneficial lactic acid producing bacteria have also been used in recent years as feed additives to promote better feed utilization in many types of animals.

DEPR:
There are various known strains of non-pathogenic lactic acid producing bacteria including some species of the genus Streptococcus and the entire genus Lactobacillus. Among the lactic acid producing species within these two generic categories are Streptococcus lactus, Streptococcus cremoris, Streptococcus diacetylactis, Streptococcus thermophillus and Streptococcus faecium. Also Lactobacillus acidophilius, Lactobacillus alimentarius, Lactobacillus bifidus, Lactobacillus brevis, Lactobacillus buchneri, Lactobacillus bulgaricus, Lactobacillus casei, Lactobacillus catenaforme, Lactobacillus cellobiosus, Lactobacillus collinoides, Lactobacillus confusus, Lactobacillus curvatus, Lactobacillus delbrueckii, Lactobacillus farciminis, Lactobacillus fermentatae, Lactobacillus fermentum, Lactobacillus fructivorans, Lactobacillus fructosus, Lactobacillus helveticus, Lactobacillus heterohiochi, Lactobacillus hilgardii, Llactobacillus homohiochi, Lactobacillus jensenii, Lactobacillus lactis, Lactobacillus leichmannii, Lactobacillus malefermentans, Lactobacillus mali, Lactobacillus maltaromicus, Lactobacillus minutus, Lactobacillus pentoaceticus, Lactobacillus plantarum, Lactobacillus rogosae, Lactobacillus ruminis, Lactobacillus sake, Lactobacillus lalivarius, Lactobacillus sanfrancisco, Lactobacillus thermophilus, Lactobacillus trichodes, Lactobacillus viridescens, Lactobacillus vitulinus, and Lactobacillus xylosus.

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L9: Entry 4 of 18 File: USPT Dec 15, 1998

US-PAT-NO: 5849289
DOCUMENT-IDENTIFIER: US 5849289 A

TITLE: Method for inhibiting microorganism growth

DATE-ISSUED: December 15, 1998

INVENTOR-INFORMATION:

| NAME | CITY | STATE | ZIP CODE | COUNTRY |
|---|---|---|---|---|
| Dobrogosz; Walter J. | Raleigh | NC | | |
| Lindgren; Sven E. | Uppsala | | | SEX |

ASSIGNEE-INFORMATION:

| NAME | CITY | STATE | ZIP CODE | COUNTRY | TYPE CODE |
|---|---|---|---|---|---|
| Biogaia Biologics AB | Gothenburg | | | SEX | 03 |

APPL-NO: 8/ 476630
DATE FILED: June 7, 1995

PARENT-CASE:
This is a divisional application of Ser. No. 084/214,014, filed Mar. 16, 1994, (U.S. Pat. No. 5,439,678), which is a continuation of Ser. No. 07/708,800 filed May 30, 1991 ( (now abandoned), which is a continuation of Ser. No. 07/268,361 filed Sep. 19, 1988 (now abandonded), which is a continuation-in-part of Ser. No. 07/102,830 filed Sep. 22, 1987 (now abandoned), which is a continuation-in-part of Ser. No. 07/046,027 filed May 1, 1987 (now abandoned).

INT-CL: [6] A01N 63/00, C12N 1/20
US-CL-ISSUED: 424/93.45; 426/61, 514/693, 435/123, 435/252.1, 435/34, 435/244
US-CL-CURRENT: 424/93.45; 426/61, 435/123, 435/244, 435/252.1, 435/34, 514/693
FIELD-OF-SEARCH: 424/93.45, 435/123, 435/124, 435/252.1, 435/34, 435/244, 426/61, 514/693

PRIOR-ART-DISCLOSED:

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L9: Entry 4 of 18 File: USPT Dec 15, 1998

DOCUMENT-IDENTIFIER: US 5849289 A
TITLE: Method for inhibiting microorganism growth

BSPR:
The metabolic endproducts of Lactobacillus metabolism such as acetic acid, lactic acid and hydrogen peroxide are well-known for their antimicrobial activities. Two laboratories have reported that the heterofermentative species Lactobacillus brevis, Lactobacillus buchneri (8) and Lactobacillus strain 208-A (9,10) metabolize glycerol anaerobically. The latter strain carries out an anaerobic dehydration (involving glycerol dehydratase) of 2 moles of glycerol yielding 2 moles of .beta.-hydroxypropionaldehyde which in turn is dismutated to 1 mole of .beta.-hydroxypropionic acid and 1 mole of 1,3-propanedlol. Some lactobacilli also produce bacteriocins or bacteriocin-like proteins which exhibit bacteriocidal activity toward other members of that species or closely related species. Some unconfirmed reports have appeared concerning low molecular weight, antimicrobial substances produced by lactobacilli. Although their existence has been predicted for some time, such substances have not been confirmed or isolated.

BSPR:
According to the present invention, biologically pure strains of L. reuteri are provided. Under the controlled cultivation methods of the invention, these strains produce a newly isolated and characterized broad-spectrum antimicrobial substance termed reuterin. This antibiotic may be used to kill other microorganisms under defined conditions using a microorganism (L. reuteri) that is nonpathogenic to humans and other animals. The technique of the invention for isolation of reuterin-producing Lactobacillus reuteri strains may also be used to isolate strains from humans and agriculturally important animals so that these isolated strains may be used as probiotic agents for the specific animal from which they were isolated. Thus, L. reuteri 1063, isolated from swine has potential use as a probiotic agent in moderating colibacillosis and weanling diarrheal disease in swine and for increasing their feed efficiencies. In comparison to a number of other homo- and heterofermentative lactobacillil isolated directly from swine small intestines, and also in comparison to L. reuteri strains 20016 and 27273 which have been held in stock culture for long periods of time, L. reuteri 1063 demonstrates strong auto-agglutination, a high degree of surface hydrophobicity and binds better than other strains to swine epithelial cells in culture. A process for the production of reuterin and a procedure for isolation of reuterin-producing strains of L. reuteri from the GI tract (or stools) of all animals harboring this species are also provided. Production of large quantities of a naturally occurring broad spectrum antibiotic as provided by the invention makes possible the use of this antibiotic for treatment of a variety of diseases and as a general purpose antimicrobial agent.

US 0976735507P1

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